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Patentanmeldung Nr. | Patent application No. | Demande de brevet n°

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Process and intermediate useful in the preparation of vitamin B6

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Process and intermediate useful in the preparation of vitamin B_6

The present invention relates to a process useful in the preparation of vitamin B_6. More particularly, the present invention relates to the preparation of N-alkoxyoxalyl alanine alkyl esters (hereinafter : EOAE). Furthermore, the invention also relates to novel intermediates occuring in the process of the invention, N-alkoxyoxalyl alanines (hereinafter: EOA).

There have been numerous reports, patents and publications for the preparation of ethoxyoxalyl alanine alkyl esters. The majority of these are two step approaches which involve first an esterification of alanine under acidic conditions (typically via alanine hydrochloride) in the appropriate alcohol. This is followed by neutralization of the ammonium salt and amide bond formation with ethoxalyl chloride or diethyloxalate, see, e.g., Chem. Ber. 30 (1897), 579; Bull. Chem. Soc. Jap. 42(5) (1969), 1435-7; and patent publications, FR 1 533 817 and JP 43010614.

A one step approach to prepare EOAE directly from alanine is disclosed in FR 2 010 601. Here in the examples, importantly, when diethyloxalate was generated *in situ* with excess oxalic acid (3 eq.) in ethanol, in addition to the desired EOAE, *N*-formyl alanine ethyl ester, (NFAE), was produced as a by-product in up to 10 % under these conditions. The research group later disclosed a modified one step approach in Bull. Chem. Soc. Jpn. 45, (1972), 1917-1918, which minimized the formation of *N*-formyl alanine ethyl ester to trace levels. This synthesis combined alanine with 1 eq. oxalic acid and 5 eq. diethyloxalate. The reaction was performed using ethanol as the solvent in an autoclave at 120° C under repeated heating cycles and afforded EOAE in up to 70 % yield. Importantly, when diethyloxalate was generated *in situ* with excess oxalic acid (3 eq.) in ethanol, in addition to

the desired EAOE, *N*-formyl alanine ethyl ester, (NFAE), was produced as a by-product in up to 10 % under these conditions. At higher temperatures the yield of NFAE increased remarkably.

Another one-pot approach under acidic conditions is disclosed in the patent publications CN 86101512 and CN 87100359 A. Here for the esterification step, alanine was reacted with oxalic acid in an aqueous HCl-EtOH solvent mixture. Benzene was added to assist in azeotropic removal of the formed water. Finally, diethyloxalate and anhydrous sodium carbonate were added to the crude ester product. After the one-pot reaction, the following steps were necessary: 1) distillation of the low boiling components, 2) addition of water to dissolve remaining solids, 3) separation of the organic phase, 4) extraction of the aqueous phase, 5) final distillation of the combined organic phases to recover solvents, excess diethyloxalate and to isolate the desired product EOAE (reported yield from alanine 88%). These required multiple work-up steps are tremendous disadvantages in this one-pot approach.

The process of the present invention allows the preparation of N-alkoxyoxalyl alanine alkyl ester from alanine in high yield while avoiding the drawbacks inherent to known processes. Particularly, the process of the present invention avoids the generation of large amounts of inorganic salt waste normally found in the known acidic approaches to EOAE.

Thus, in one aspect the invention relates to a process for the preparation of N-alkoxyoxalyl alanine alkyl ester which comprises reacting alanine with an alkyl, preferably a lower alkyl, oxalate under substantially non-acidic conditions.

The term alanine as used herein comprises racemic (D,L)-alanine as well as the enantiomers, L- and D-alanine, and mixtures thereof in any ratio. To establish substantially non-acidic conditions a base may be added to the reaction mixture. In a preferred aspect, the reaction is carried out without the addition of a base. If a base is added, it is preferably an organic base, particularly an organic base of the formula NR_3 where R is lower alkyl, such as triethylamine, or is a cyclic tertiary amine, such as pyridine, quinoline, N-methyl pyrrolidine or N-methyl piperidine, or tertiary amide, such as N-methyl pyrrolidone, or mixtures thereof. The term "lower alkyl" as used herein refers to linear or branched alkyl moieties containing 1-8, preferably 1-4 carbon atoms. The most preferred lower alkyl moiety is ethyl.

The molar ratio of alanine to oxalate is suitably from about 1:2 to about 1:10, preferably from about 1:3 to about 1:6. The reaction is suitably carried out at a temperature from about 120 °C to about 200 °C, preferably from about 135 °C to about 160 °C.

In a preferred embodiment of the present invention, the reaction is carried out at atmospheric pressure while ensuring that substantially all of the ethanol produced during the reaction remains in the reaction system. This can be accomplished e.g., by carrying out the reaction in an autoclave (i.e., at elevated pressure), or, more preferably, under atmospheric pressure with efficient cooling of the vapour phase of the reaction mixture, thus causing the alcohol formed in the reaction to be retained in the reaction system. According to that embodiment of the process of the invention, the reaction of alanine with the oxalate, preferably, diethyl oxalate, is carried out for about 8 hrs. at about 145 °C to about 150 °C with the molar ratio of alanine to oxalate being about 1:4. In still another embodiment of the process of the invention, a lower alkanol, preferably one corresponding to the alkyl moiety of the oxalate used in the reaction, can be added.

If the reaction is carried out under atmospheric pressure in the presence of an organic base the reaction is suitably performed by heating the reaction mixture for about 4-12 hrs, preferably about 6-10 hrs, to a temperature below the boiling point of the organic base. During heating, the temperature is suitably from about 60 °C to about 160 °C, preferably from about 80 °C to about 120 °C, most preferably from about 90 °C to about 110 °C. Preferably, a low-boiling base, i.e. a base having a boiling point substantially below about 135 °C under atmospheric pressure is used. Thereafter, any low-boiling organic base is removed from the reaction mixture, e.g., by distillation and the reaction temperature is then increased up to about 160 °C for a duration sufficient to complete the formation of the desired product, EOAE.

It has been found that in the process of the present invention an N-alkoxyoxalyl alanine may occur as an intermediate. N-alkoxyoxalyl alanines (EOA) such as N-ethoxyoxalyl alanine compoundare novel compounds and as suchis also an object of the present invention. They can be readily converted into EOAE by heating in the presence of a dialkyl oxalate such as diethyl oxalate and can be isolated from the reaction mixture formed in the process of the invention. Thus, the process of the present invention can be carried out with or without isolation of EOA. When the one-pot EOAE synthesis is

performed using NEt_3, EOA is generated as the major product after the first heating sequence. These conditions may suitably be used to produce EOA in major quantity.

A so-obtained N-alkoxyoxalyl alanine alkyl ester can be converted by known methods into vitamin B_6, e.g. by cyclisation to form 4-methyl-5-alkoxy (e.g.,ethoxy)-2-alkoxy (e.g.,ethoxy) carbonyl-oxazole which on treatment with alkali and, subsequently, acid is converted into 4-methyl-5-ethoxyoxazole.4-methyl-5-alkoxy (e.g.,ethoxy) oxazole. The latter, on reaction with4,7-dihydro-2- 4,7-dihydro-2-isopropyl-1,3-dioxepin can be further converted into vitamin B_6 as disclosed in FR 1 533 817 and US 3,250, 778 the contents of which are incorporated herein by reference.

The invention is illustrated further by the Examples. EOA denotes N-ethoxyoxalyl alanine. GC means gas chromatography [GC column RESTEK, Rtx-5SilMS, 30 m, 0.28 micron], EOA was isolated by column chromatography [Merck Silica gel 60 (0.040-0.063 mm)].

Example 1:

To a reaction flask under a stream of argon there was added D,L-alanine (17.9 g, 200 mmol) followed by triethylamine (24.4 g, 33.4 ml, 240 mmol, 1.2 eq) and last diethyloxalate (117.5 g, 800 mmol, 4 eq). A gas trap containing 100 mL of a 10 % aqueous $BaCl_2$ solution was connected to the reactor's gas outlet in order to trap any CO_2 produced during the reaction. The reaction mixture was heated at 105° C (internal temp) for 8 h (after 6 h all alanine was in solution). GC analysis at this point in the reaction sequence showed 1.2 w/w % N-ethoxyoxalyl alanine ethyl ester and 22.3 w/w% N-ethoxyoxalyl alanine as the major products (98.4 % yield based on alanine). No CO_2 production was observed during the first heating phase. The reaction mixture was then heated up to 145° C over 30 min and heated a further 8 h. During this second heating, triethylamine and ethanol were distilled into a collector. In the second heating phase, CO_2 production occurred, $BaCO_3$ precipitate was observed in the gas trap. GC analysis of the reaction mixture at the end of the reaction showed N-ethoxyoxalyl alanine ethyl ester produced as the major product in 28.7 GC w/w% with some EOA remaining 2.2 GC w/w%. Overall yield (EOAE(N-ethoxyoxalyl alanine ethyl ester + N-ethoxyoxalyl alanine) of the 2 step, one pot process calculated from alanine was 73.9 %.

Example 2:

To a stainless steel autoclave under an argon atmosphere there was added D,L-alanine (0.9 g, 10 mmol) followed by triethylamine (1.2 g, 1.7 ml, 12 mmol, 1.2 eq.) and diethyloxalate (5.9 g, 40 mmol, 4 eq). The reaction mixture was heated at 150° C (heating block

temperature) for 8 h. The internal pressure following the reaction was circa 5 bar. The autoclave was allowed to cool over 15 min and a sample of the brownish reaction mixture was removed for GC analysis. The reaction mixture contained N-ethoxyoxalyl alanine ethyl ester as the major product in 22.0 GC w/w% and some EOA 0.5 GC w/w%. The overall yield (EOAE(N-ethoxyoxalyl alanine ethyl ester + N-ethoxyoxalyl alanine) based on alanine was 79.2 %.

Example 3:

To a reaction flask under a stream of argon there was added D,L-alanine (8.9 g, 100 mmol) followed by diethyloxalate (58.8 g, 400 mmol, 4 eq). A gas trap containing 100 mL of a 10 % aqueous $BaCl_2$ solution was connected to the reactor's gas outlet in order to trap any CO_2 produced during the reaction. The reaction mixture was heated at 145° C (internal temp) for 7 h (after 6 h all alanine was in solution). GC analysis of the reaction mixture at the end of the reaction showed N-ethoxyoxalyl alanine ethyl ester produced in 12.5 GC w/w% with N-ethoxyoxalyl alanine remaining as the major product 16.0 GC w/w%. Overall yield (EOAE(N-ethoxyoxalyl alanine ethyl ester + N-ethoxyoxalyl alanine) of the 2 step, one pot process calculated from alanine was 95.0 %.

Example 4:

To a stainless steel autoclave under an argon atmosphere there was added D,L-alanine (2.23 g, 25 mmol) followed by diethyloxalate (14.69 g, 100 mmol, 4 eq). The reaction mixture was heated at 150° C (heating block temperature) for 8 h. The internal pressure following the reaction was circa 5 bar. The autoclave was allowed to cool over 15 min. The unreacted alanine was removed by filtration (1.5 g, 17.3 mmol; 69.1 % recovered) and a sample of the light brown reaction mixture was removed for GC analysis. The reaction mixture contained N-ethoxyoxalyl alanine ethyl ester 1.0 GC w/w% and N-ethoxyoxalyl alanine 3.0 GC w/w% as the major product. The overall yield (EOAE(N-ethoxyoxalyl alanine ethyl ester + N-ethoxyoxalyl alanine) based on alanine was 26.5 %. The corrected yield of the reaction based on recovery of the unreacted alanine was 95.6 %.

Example 5:

Apparatus: 500 mL double jacketed 4 neck glass reactor with circulation thermostat temperature control (Julabo FP50-MH), IKA Eurostar Digi-Visc stirrer with Ekato-Intermig propeller, Pt 100 thermometer/controller, Dephlegmator, reflux cooler, vacuum system – Vakuubrand with CVC2 vacuum controller, cold traps, and argon degassing.

To the double jacketed reactor under an argon atmosphere there was added D,L-alanine (44.7 g, 500 mmol) followed by diethyloxalate (298.6 g, 2000 mmol, 4 eq.). The suspension was stirred at 200 rpm and during 1 h was heated up to an internal temperature of 145° C (Mantel temperature 150° C). At the start of the reaction the dephlegmator was set to 5° C. Alanine was completely dissolved after circa 6 h reaction time. During the course of the reaction, the internal temperature dropped from 145° C to 115° C. After 26 h, the dephlegmator was heated to 70° C and ethanol and other low boiling compounds were distilled away from the reaction over 30 min. at internal temperature of 116° C and a vacuum from 900 mbar to 400 mbar into a prefraction collection flask. The collection flask was changed and the dephlegmator heated to 110° C. The unreacted excess diethyloxalate was distilled away from the reaction products over 2 h, internal temperature from 116° C to 154° C and under a vacuum from 400 mbar to 40 mbar. The reaction mixture containing the crude desired product was cooled to room temperature, analyzed by GC, and found to contain 77.0 w/w % N-ethoxyoxalyl alanine ethyl ester and 5.5 w/w% EOA. The overall yield EOA)(EOAE(N-ethoxyoxalyl alanine ethyl ester + N-ethoxyoxalyl EOA)alanine) based on alanine was 75.0 %.

Example 6:

To a stainless steel autoclave under an argon atmosphere there was added D,L-alanine (0.9 g, 10 mmol) followed by ethanol (1.8 g, 2.3 ml, 40 mmol, 4 eq.) and diethyloxalate (5.9 g, 40 mmol, 4 eq). The reaction mixture was heated at 150° C (heating block temperature) for 8 h. The internal pressure following the reaction was circa 5 bar. The autoclave was allowed to cool over 15 min and a sample of the pale yellow reaction mixture was removed for GC analysis. The reaction mixture contained N-ethoxyoxalyl alanine ethyl ester as the major product in 18.5 GC w/w% and some EOA 1.7 GC w/w%. The overall yield (EOAE(N-ethoxyoxalyl alanine ethyl ester + N-ethoxyoxalyl alanine) based on alanine was 78.4 %.

Example 7:

To a flask under an atmosphere of argon there was added alanine (8.9 g, 100 mmol) followed by triethylamine (12.2 g, 8.9 ml, 120 mmol, 1.2 eq.) and diethyloxalate (59.7 g, 400 mmol, 4 eq.). The reaction mixture was heated at 100° C for 7 h. The mixture was cooled to 2-3° C and stirred a further 2 h. The unreacted alanine that precipitated from the reaction mixture was isolated by filtration (Sartorius-Membrane Filter, reg. Cellulose, 0.45 microns). The triethylamine was first removed by rotary evaporation (to 20 mbar, 60° C), then the excess diethyloxalate was removed under high vacuum rotary evaporation (to 0.01 mbar, 70° C). The remaining crude product (14.5 g), a yellow-gold oil, was analyzed by

GC. N-ethoxyoxalyl alanine was found as the major product (80.2 w/w% GC) in a yield based on alanine of 62 %. For isolation and characterization of N-ethoxyoxalyl alanine the crude product was purified by column chromatography [Merck silica gel 60 (0.040-0.063 mm), EtOAc/CH_3CN 8:2 v/v, 2 % HOAc), column size (8 x 20 cm)] to yield 1.0 g of pure EOA (>99 GC w/w%) as an off-white solid and 9.0 g mixed fractions (N-ethoxyoxalyl alanine + monoethyloxalate, which were not efficiently separated under the conditions). The purity of the isolated N-ethoxyoxalyl alanine was high enough to allow for full characterization of the structure (see below).

Structure Data:

$C_7H_{11}NO_5$ (189.17): *N*-ethoxyoxalyl alanine

R_f (SiO_2, EtOAc/CH_3CN 8:2 v/v, 2 % HOAc) = 0.30.

mp = 73-74° C (uncorrected).

^{1}H-NMR (400 MHz, DMSO-d_6): δ = 1.28 (t, J = 7.0 Hz, 3 H, OCH_2CH_3), 1.34 (d, J = 7.2 Hz, 3 H, CH_3), 4.24 (q, J = 7.0 Hz, 2 H, OCH_2CH_3), 4.26 (dq, J = 7.6, 7.2 Hz, 1 H, $NHCHCH_3$), 9.02 (d, J = 7.6 Hz, 1 H, CONHCH), 12.70 (bs, 1 H, COOH).

^{13}C-NMR (100 MHz, DMSO-d_6): δ = 13.8, 16.5, 47.9, 62.0, 156.9, 160.5, 172.9.

IR (KBr): ν = 3365, 3126, 3006, 1733, 1664, 1539, 1278, 1193 cm^{-1}.

MS (elec. Spray, neg. mode): 188.2 $(M-H)^-$

CHN: calc. C 44.45, H 5.86, N 7.40, O 42.29; found C 44.20, H 5.78, N 7.11, O 42.58.

What is claimed is :

1. Process for the preparation of N-alkoxyoxalyl alanine alkyl esters which comprises reacting alanine with a di-alkyl oxalate under substantially non-acidic conditions.

2. A process as in claim 1 wherein the molar ratio of alanine to oxalate is from about 1:2 to about 1:10, preferably from about 1:3 to about 1:6.

3. A process as in claim 1 or 2 wherein the reaction temperature is from about 120 °C to about 200 °C, preferably from about 135 °C to about 160 °C.

4. A process as in any one of claims 1-3 wherein the reaction is carried out at atmospheric pressure while ensuring that substantially all of the alkanol produced remains in the reaction system.

5. A process as in any one of claims 1-3 wherein the reaction is carried out at elevated pressure.

6. A process as in claim 5 wherein a lower alkanol, preferably ethanol, is added.

7. A process as in claim 6 wherein the alkyl moiety of the lower alkanol corresponds to the alkyl moiety in the oxalate ester group.

8. A process as in claim 1 wherein alanine is reacted with a di-lower alkyl oxalate for about 8 hours at about 135 °C to about 160 °C.

9. A process as in claim 8 wherein the molar ratio of alanine to oxalate is about 1:4.

10. A process as in claim 1 wherein the reaction is carried out in the presence of a base.

11. A process as in claim 10 wherein the base is an organic base.

12. A process as in claim 11 wherein the organic base is of the formula NR_3 where R is lower alkyl, or is a cyclic tertiary amine or tertiary amide, or mixtures thereof.

13. A process as in claim 12 wherein the organic base is triethylamine, pyridine, quinoline or N-methyl pyrrolidone.

14. A process as in claim 12 wherein the organic base is triethylamine.

15. A process as in any one of claims 10-14 wherein the molar ratio of alanine to oxalate is from about 1:2 to about 1:10, preferably from about 1:3 to about 1:6.

16. A process as in any one of claims 10-15 wherein the reaction temperature is in a first step from about 60 °C to about 160 °C, preferably from about 80 °C to about 120 °C, most preferably from about 90 °C to about 110 °C, whereupon any low boiling organic base is removed from the reaction mixture and the reaction temperature increased, in a second step, up to about 160 °C.

17. A process as in claim 16 wherein the reaction time in the first step is about 4-12 hrs, preferably about 6-10 hrs, and in the second step about 4-12 hrs., preferably about 6-10 hrs.

18. A process for the preparation of vitamin B_6 which comprises preparing a N-alkoxyoxalyl alanine alkyl ester in accordance with any one of claims 1-17 and converting it by known methods into vitamin B_6.

19. A process as in any one of the preceding claims wherein the alkyl moieties are lower alkyl moieties.

20. N-alkoxyoxalyl alanines.

21. N-ethoxyoxalyl alanine.

22. The invention as described hereinbefore, especially with reference to the Examples.

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